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Nutritional Composition of Grain Legume Leaves and the Impact of Leaf Removal on Yield

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Abstract

Grain legumes can thrive in adverse environments, making them a climate-smart technology for hunger mitigation. Although several countries rely immensely on grain legumes to meet daily protein intake requirements per capita, the potentiality of leaf utilization for protein and other nutrients has not been widely considered; additionally, insufficient information is available on leaf removal effects on yield and leaf nutritional composition of grain legumes. A 2-yr experiment was conducted in central Iowa, USA, to determine the effects of leaf removal rates on nutritive value of removed leaf tissue and subsequent grain yield of cowpea [*Vigna unguiculata* (L.) Walp.], lablab [*Lablab purpureus* (L.) Sweet], and soybean [*Glycine max* (L.) Merr.]. Across entries, dry leaf mean nutrient concentration was 229 g kg^{-1} for CP, and 17,832, 4461, 21,991, 3702, 113, 205, and 86 mg kg^{-1} for Ca, Mg, K, P, Mn, Fe, and Zn, respectively. Yield and major yield attributes were affected by leaf removal rate in 2014, but not in 2013. In 2014, grain legumes with 0% leaf removal had 20, 32, and 35% greater yield and seeds weighed 6, 11, and 12% more than those with 33, 66, and 99% leaf removal, respectively. Aboveground biomass, yield, and yield components also differed among entries both years. Grain legume leaf utilization as vegetable or forage may improve human and ruminant nutrition by using leaves, especially in developing countries.

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Comments

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Nutritional Composition of Grain Legume Leaves and the Impact of Leaf Removal on Yield

Rosemary Bulyaba* and Andrew W. Lenssen

Core Ideas

- Soybean, cowpea, and lablab leaves differed in nutritive value.
- Soybean and cowpea grain yield responded differently to leaf harvesting done at Vegetative Stage 6 (V6).
- Legume plants from which two leaves (66%) are harvested at V6 may have similar grain yield to those whose leaves are not harvested.

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ABSTRACT

Grain legumes can thrive in adverse environments, making them a climate-smart technology for hunger mitigation. Although several countries rely immensely on grain legumes to meet daily protein intake requirements per capita, the potentiality of leaf utilization for protein and other nutrients has not been widely considered; additionally, insufficient information is available on leaf removal effects on yield and leaf nutritional composition of grain legumes. A 2-yr experiment was conducted in central Iowa, USA, to determine the effects of leaf removal rates on nutritive value of removed leaf tissue and subsequent grain yield of cowpea [*Vigna unguiculata* (L.) Walp.], lablab [*Lablab purpureus* (L.) Sweet], and soybean [*Glycine max* (L.) Merr.]. Across entries, dry leaf mean nutrient concentration was 229 g kg^{-1} for CP, and 17,832, 4461, 21,991, 3702, 113, 205, and 86 mg kg^{-1} for Ca, Mg, K, P, Mn, Fe, and Zn, respectively. Yield and major yield attributes were affected by leaf removal rate in 2014, but not in 2013. In 2014, grain legumes with 0% leaf removal had 20, 32, and 35% greater yield and seeds weighed 6, 11, and 12% more than those with 33, 66, and 99% leaf removal, respectively. Aboveground biomass, yield, and yield components also differed among entries both years. Grain legume leaf utilization as vegetable or forage may improve human and ruminant nutrition by using leaves, especially in developing countries.

Abbreviations: ADF, acid detergent fiber; ADL, acid detergent lignin; CP, crude protein; ICP-OES, inductively coupled plasma–optical emission spectrometer; LAI, leaf area index; NDF, neutral detergent fiber; NIRS, near infrared spectroscopy; PLS, pure live seeds; QTL, quantitative trait loci; RM, relative maturity.

The increasing demand for protein to meet human and livestock nutritional requirements means inexpensive, sustainable sources are needed. In the livestock industry, supplementary pastures are essential for economical production, especially in regions with inadequate permanent pastures or dry seasons with poor forage productivity (Gibson et al., 1943). Additionally, there is great public health concern about protein, energy, and micronutrient deficiencies such as Fe, Zn, I, and vitamin A in developing countries, especially those in South Asia and sub-Saharan Africa (FAO, 2004; Muller and Krawinkel, 2005). Dietary diversification through grain legume leaf utilization may be a management practice to provide nutritional supplementation to alleviate deficiencies.

In Asia and Africa, consumption of young, tender leaves by humans and utilization of older leaves as forage is common for grain legumes such as lablab [*Lablab purpureus* (L.) Sweet], also known as hyacinth bean, and cowpea [*Vigna unguiculata* (L.) Walp.] (Saidi et al., 2007; Baloyi and Ayodele, 2013). Cowpea is among the top four leafy vegetables in many African countries (Barrett, 1990). In Botswana, for instance, legume vegetable green leaves are harvested throughout the growing season at all stages of development, although the effect of leaf harvesting on yield is not well documented (Demooy and Demooy, 1989). Farmers lack sufficient knowledge about the proportion of leaves to be harvested and at what growth stages leaves may be harvested without affecting grain yield (Badi et al., 2012).

Grain legume leaf removal has variable impact on aboveground biomass, grain yield, and grain nutritional composition. Teigen and Vorst (1975) reported that leaf removal increased soybean [*Glycine max* (L.) Merr.] aboveground dry matter accumulation by enabling more light to reach lower leaves, which increased plant photosynthetic rate. By reducing competition for resources, leaf removal may increase the efficiency of remaining leaves. Weber (1955) reported that due to increased efficiency of the remaining leaves, yield was marginally reduced. This was reported at 10% leaf removal (including half of leaf tissue/leaflet from every node and the trifoliate leaflets) done between when soybean plants had five to six unrolled trifoliate leaves and less than 1% flowers. However, studies with cowpea found that grain yield was decreased following leaf removal, perhaps due to altered source–sink relations when immature trifoliates were harvested at 7-d intervals at 2, 4, and 5 wk after emergence (Saidi et al., 2007). Additionally, leaf removal and frequency decreased total biomass and biomass partitioning to roots, stems, leaves, pods, and grain size in soybean, cowpea, and lablab, especially when done during the reproductive stages(Enyi, 1975; Wood, 1983 Demooy and Demooy, 1989). According to the authors, this may be attributable to decreased N fixation and available N for seed formation.

Leaf removal can influence various yield components. Board and Harville (1993) reported that soybean seed and pod number declined with leaf removal, whereas a decrease in seed weight and size following defoliation were reported by Egli and Leggett (1976) and Fehr et al. (1981). These differences may be attributed to the developmental stage at which leaf removal was done (Board et al., 1994). Board and Harville (1993) found that leaf removal during early reproductive stages primarily decreased pod number in soybean. Enyi (1975) emphasized the importance of growth and development stage at which leaf removal was done in studies on legumes—groundnut (*Arachis hypogea* L.), cowpea, and soybean. He explained that during early vegetative and reproductive stages, assimilates produced by leaves were mainly used for development of main/primary stems to new leaves and pod initiation and growth, respectively; therefore, leaf removal during vegetative stages reduced weight of side/secondary and main/primary stems, number of side/secondary branches, and plant height. Barrett (1987) further reported that timing of leaf removal affected cowpea ability to recover and produce high grain yield. He emphasized that removal of too many young leaves decreased seed yield, whereas removal of the oldest leaves increased grain yield. Additionally, stages between R4 (full pod stage when seed development begins), R5 (seed filling stage and dry weight seed accumulation), or R5.5 (between beginning of seed fill and full pod development) were reported to be sensitive to defoliation, leading to 80% soybean grain yield reduction at 100% defoliation (Goli and Weaver, 1986; Board et al., 1994). Board et al. (1994) explained that yield sensitivity to leaf removal increased as seed filling progressed. The authors added that plant response to lower leaf area index (LAI) and light interception decreased pod number in soybean, although seeds per pod and seed weight were unaffected.

Crop response to leaf removal may be influenced by several factors, such as cultivar, leafing intervals/frequency, position of harvested leaves on the plant, and leaf harvesting percentage. In a study with lablab, plant cuttings similar to leaf removal done at heights of 10 and 20 cm between 12 and 18 wk after planting led to lower seed yield compared with cuttings done at maturity (Ogedegbe et al., 2012). Saidi et al. (2007) also reported that, both time from crop emergence to first leaf harvest in addition to leaf harvesting frequency impacted yield in cowpea. They reported that weekly leaf removal gave a higher leaf yield, but lowered grain yield compared to biweekly removal; grain yield was lowest when leaf removal initiated in the second week after emergence. Grain yield was greater when leaf removal delayed to the fifth week after emergence. Working with cowpea, Karikari and Molatakgosi (1999) reported that grain yield following leaf removal was dependent on both leaf harvesting intensity and cultivar. They found that 50% leaf removal increased grain yield, whereas 75% leaf removal decreased yield. Johnston and Pendleton (1968) reported that the position of harvested leaves influenced the plant's response to leaf removal. They reported that removal of upper, newer leaves caused a 17% reduction in soybean grain yield compared with 4 and 22% yield reductions after the bottom and middle leaf removal, respectively. Overall, it is apparent that leaf removal from grain legumes in reproductive phases has a substantial negative influence on grain yield.

Few reports exist on the nutritive value of cowpea, soybean, and lablab leaves harvested at the sixth trifoliate leaf stage (V6) (Schwartz and Langham, 2010; Licht 2014) and the potential that utilization of these leaves as leafy vegetables may have in supplementing nutrition. Macro and micronutrient deficiencies are prevalent in many developing countries. The latter such as Fe, Zn, and vitamin A, contributing to long-term causal relationships between nutritional deficiencies, work productivity and physical activity such as impaired cognitive development and tissue oxidative capacity impairment, increased hospitalizations and disability among others (WHO, 2017). Utilization of legume leaves may supplement some macro and micronutrients into high-carbohydrate diets to alleviate nutritional deficiencies. Because the influence of leaf removal on nutritional composition of harvested leaves from soybean, cowpea, and lablab at V6 stage is not well documented, we conducted a study to (i) determine the nutritional composition of soybean, cowpea, and lablab leaves harvested at V6, and (ii) determine the influence of leaf harvesting at V6 on subsequent biomass and grain yield.

MATERIALS AND METHODS

Field studies in 2013 and 2014 were conducted in Ames and Boone, IA, respectively. In 2013, the field trials were located at the Iowa State University Curtiss Farm (42°00′18.0″ N, 93°40′09.3″ W). In 2014, field trials were located at the Agricultural Engineering and Agronomy Research farm (42°1′18.76″ N, 93°46′35.94″ W). Both farms are tile and ditch drained due to poor natural drainage and excess water in the area (Hofstrand, 2010). Soil samples were collected from 0 to 30 cm before planting. The soil samples were then analyzed for pH using a standard Fisher pH and electrodes (Watson and Brown, 1998), organic matter by dry combustion (Combs and Nathan, 1998), available P and K (Mehlich-3) (Warncke and Brown, 1998), and nitrate (Gelderman and Beegle, 1998) at the Soil and Plant Analysis Laboratory, Iowa State University (Table 1). The predominant soils at the Curtiss farm were Canisteo clay loam (fine-loamy, mixed, superactive, calcareous, mesic Typic Endoaquolls) (NCSS, 2015a) and Nicollet loam (fine-loamy, mixed, superactive, mesic Aquic Hapludolls) (NCSS, 2015c) (Table 1). The soils at the Agricultural Engineering and Agronomy Research farm were predominantly Canisteo silty clay loam and Clarion loam (fine-loamy, mixed, superactive, mesic Typic Hapludolls) (NCSS, 2015b), and Bemis moraine (loamy, mixed, active, acid, shallow Aeric Cryaquepts) soils (NRCS, 2014) (Table 1). Both sites had been in a corn (*Zea mays* L.)–soybean rotation for several decades, with no known production history of cowpea or lablab.

The experiment was a complete factorial of pulse grain crops and four leaf removal rates in a randomized complete block with three replications. In 2013, the three grain legumes used were 'Iron

Table 1. Preplant soil test values for Story County (2013) and Boone County (2014) experimental sites in Iowa.

Year	Depth	Mehlich 3-P	Mehlich 3-K	pH	OM	NO_3
	cm	———mg kg^{-1}	———		g kg^{-1}	mg kg^{-1}
2013	0–15	35	198	5.7	4.6	4
2013	15–30	7	136	6.5	4.2	5
2014	0–15	43	153	5.5	3.6	7
2014	15–30	14	99	5.6	2.7	4

and Clay' cowpea, 'Rongai' lablab, and 'P92Y82' soybean (DuPont Pioneer, Johnston, IA) (2.8 relative maturity, RM). In 2014, the four grain legumes used were two cowpea cultivars, 'CA46' California black-eyed pea and 'Top Crop' purple hull pinkeye southern pea, Rongai lablab, and P92Y82 soybean. The leaf removal rates at vegetative stage six (V6) were 0, 33, 66, and 99% corresponding to 0, 2, 4, and 6 leaves removed/harvested from the top of each plant. Each plot consisted of four rows and individual plot size was 7.6 m long by 3 m wide with 0.76-m row spacing. The seeding rate was 34 pure live seeds (PLS) m^{-2} and plots were planted on 13 June 2013 and 6 June 2014. Seeds were planted at a depth of 2.5 cm using a Maxemerge planter (John Deere, Moline, IL).

Fertilizers were not applied at either site in 2013 or 2014 because pre-plant soil test analysis indicated adequate amounts of recommended nutrients for soybean production in Iowa (Mallarino et al., 2013) (Table 1). A pre-plant application of Prowl H_2O herbicide (BASF Ag Products) pendimethalin (*N*-(1-ethylpropyl)-3,4-dimethyl-2,6-dinitrobenzenamine) was applied at a rate of 1.92 kg a.i. ha^{-1} in 187 L H_2O ha^{-1} in 2013 and 2014. Additional weed management later in the season was mechanically accomplished by cultivation and hand hoeing. In 2013, a weed infestation later in the season in soybean was controlled by using glyphosate [*N*-(phosphonomethyl)glycine] at 3.36 kg a.i. ha^{-1} in 93 L H_2O ha^{-1}. Lambda-cyhalothrin ([1α(S*),3α(Z)]-(±)-cyano-(3-phenyloxyphenyl)methyl-3-(2-chloro-3,3,3,-tricfluoro-1-propenyl)-2,2-dimethylcycloprpanecarboxylate) was used to control Japanese beetles (*Popillia japonica*) in 2013 and potato leafhoppers (*Empoasca fabae*) in 2013 and 2014 early in the growing season. The insecticide was applied at 9.08 g a.i. ha^{-1} in 93 L H_2O ha^{-1}. The experiment solely relied on natural rainfall, although the 2013 growing season had lower precipitation than 2014 (Bulyaba and Lenssen, 2017) (Table 2).

Using plants from the two middle rows of each four-row plot, phenological stages were determined using a soybean and cowpea staging system (Fehr et al., 1981; Schwartz and Langham, 2010; Licht 2014). Plant development was between V4 (fourth trifoliate leaf stage) and V6 stages of development when stand counts were taken from 5.3 m from both rows. At V6, leaves were hand harvested/removed from plants within 11.6 m^2 of the two center rows of each plot. Leaves removed from the top of the plants were placed in labelled paper bags in the shade. The samples were then placed in a forced-air oven at 60°C to dry. The leaves in the paper bags were periodically turned to prevent mold. Dried leaves were weighed and then ground through a rotary mill (Thomas Wiley mill, Model 4, Swedesboro, NJ) to pass through a 2-mm sieve.

Aboveground biomass for all the crops in the experiment was collected when the plants were at R5.5 growth stage (about 135 d from planting), prior to leaf loss, by hand-clipping 1 m row from each plot. Biomass samples were placed in a forced-air oven at 60°C until dry, weighed, and ground using an MTD chipper shredder (Model 465, Cleveland, OH). At R8 (full maturity), the grain legumes were hand-harvested for subsequent determination of yield components. Pods were hand-harvested from 2 m of row from the two central rows of each plot. The pods were counted as they were taken off each stem, and the number of pods m^{-2} calculated. Pods were then threshed with a stationary thresher (Model Almaco BT-14, Nevada, IA). Seeds were counted using a Seedburo 801 Count-A-Pak (Seedburo Equipment, Des Plaines, IL) seed counter. The seeds were oven-dried overnight at 60°C to 0% moisture and weighed.

Subsamples of the ground leaves were used to determine total N, P, K, Mg, Ca, Mn, Fe, Cu, Zn, S, and NO_3–N at Soil and Plant Analysis Laboratory, Iowa State University. The ground leaf concentrations of P, K, Mg, Ca, Mn, Fe, Cu, Zn, and S were determined using microwave assisted nitric acid digestion followed by quantification with a Spectro Ciros inductively coupled plasma–optical emission spectrometer (ICP-OES) (USEPA, 1986; Horneck and Miller, 1998). Total nitrogen (N) was determined using Leco dry combustion (Pella, 1990; Bremmer, 1996). Crude protein was calculated as N × 6.25. Nitrate-N was determined using a 2 M KCl extraction and cadmium reduction method with a Lachat QuikChem 8000 FIA+ (Mulvaney, 1996; Gelderman and Beegle, 1998). For neutral detergent fiber (NDF), acid detergent fiber (ADF), and ash analysis, additional leaf subsamples ground through a 1-mm sieve in a Thomas Wiley laboratory mill (Model 4, 3375E15, Swedesboro, NJ) were used. The NDF, ADF, and ash analysis were done using ANKOM procedures (ANKOM Technology, Macedon, NY). Soybean grain crude protein (CP), oil, and fiber analyses were done using near infrared spectroscopy (NIRS). A subsample of cowpea seeds from each plot was ground using a cyclone mill (Model

Table 2. Twelve-year monthly average air temperature and total precipitation during the 2-yr study.†

Month	2013	2014	12-yr total avg.
	Mean air temp., °C		
Apr.	8	9	11
May	16	17	17
June	21	22	22
July	23	21	24
Aug.	23	22	23
Sept.	19	17	18
Oct.	11	11	11
Avg. air temp. (Apr.–Oct.)	17	17	18
	Total precip., mm		
Apr.	148	121	98
May	180	108	136
June	26	225	122
July	26	73	115
Aug.	30	148	130
Sept.	30	138	85
Oct.	64	119	65
Cumulative precip. (Apr.–Oct.)	504	932	751

† Adapted from Bulyaba and Lenssen, 2017.

3010-80, UDY Corp., Fort Collins, CO) through a 1.0-mm sieve followed by total N determination using a colorimetric procedure (LECO, St. Joseph, MO); CP was calculated as N × 6.25.

Data were analyzed by generalized linear mixed models PROC GLIMMIX using SAS 9.4 (SAS Institute, Cary, NC). During analysis, blocks were treated as random elements in the model, whereas leaf removal rate and crop entries were treated as fixed main effects. Iron and Clay cowpea was used in 2013; however, because it had very low yield, the crop was replaced with CA46 and Top Crop cowpea in 2014, thus data for 2013 and 2014 were analyzed separately. The PDIFF procedure was used to test for differences among means where F tests were significant for main effects and their interactions. Differences between treatments are evaluated at a significance level of $P \leq 0.05$, unless otherwise stated. Linear regression was done using PROC REG for parameters that were significantly influenced by leaf removal rate.

RESULTS

Grain Yield, Aboveground Biomass, and Stand Density 2013

At maturity, stand density was similar for lablab and cowpea, but both had greater stand density than did soybean. The stand density at R8 for lablab was 35% greater than that of soybean, whereas the stand density at R8 for cowpea was 35% greater than that for soybean.

Leaf removal rate influenced the amount of harvested leaf biomass per hectare and as expected, plots at 99% leaf removal rate had the greatest leaf biomass harvested, whereas those at 33% leaf removal had the lowest leaf biomass yields (Table 3). Leaf harvesting at 99% yielded 20 and 49% more leaf biomass per hectare than 66 and 33% leaf removal. Conversely, leaf biomass yield did not differ by entry. Total aboveground biomass accumulation was not influenced by leaf removal rate or grain legume entry (Table 3).

Grain yield differed by crop but not by leaf removal rate in 2013 (Table 3). Soybean yielded 99% more grain than cowpea, whereas lablab did not produce any grain. The main effect of leaf removal rate did not impact the number of pods m^{-2}, the number of seeds pod^{-1}, the number of seeds m^{-2}, or mean seed weight (Table 3). However, each of these factors differed between soybean and cowpea.

Nutrient and Fiber Concentrations in Harvested Leaves, 2013

The influences of crop and leaf removal rate on several nutritional concentrations were significant, but their interaction (crop and leaf removal rate) was never significant. The rate of leaf removal impacted Ca concentration of harvested leaves (Table 4). Regression analysis showed that leaf Ca concentration increased with increase in leaf removal rate (r^2 = 0.963) (Table 5). The percentage of leaf removal did not significantly impact concentrations of CP, P, K, Mg, Mn, Fe, Cu, Zn, or S in the harvested leaves in 2013 (Table 4). Although these concentrations were not affected by the interaction of crop and leaf removal percentage, P, Fe, Cu, and Zn concentrations differed by crop (Table 4). Soybean leaves had the greatest concentration of CP, whereas that in Iron and Clay cowpea and lablab leaves was lower and did not statistically differ from one another. Soybean leaves had 8 and 12% greater CP than cowpea and lablab leaves, respectively. Lablab leaves had the greatest concentration of P, followed by Iron and Clay cowpea, whereas soybean leaves had the lowest; Lablab leaves had 10 and 17% greater P than cowpea and soybean leaves, respectively. The Mg concentration in soybean and cowpea leaves was similar, and both were greater than that in lablab leaves. Soybean leaves had 28% greater Mg than lablab, whereas cowpea leaves had 27% greater Mg than lablab. Calcium

Table 3. Crop stand density at V4 and R8, aboveground biomass, and yield components for P92Y82 soybean, Rongai lablab, and Iron and Clay cowpea, Ames, IA, 2013.

Treatment	Stand density V4†	Stand density R8‡	Dry leaves	Biomass	Yield	Pods	Seeds	Seeds	Seed wt.
	no. m^{-2}		kg ha^{-1}			no. m^{-2}	no. pod^{-1}	no. m^{-2}	mg $seed^{-1}$
Leafing									
0	27	27	0	4483	1203	452	2.6	728	118
33	27	30	157 b§	4844	1233	483	3.3	771	121
66	28	24	247 ab	4402	946	395	4.0	588	123
99	29	28	307 a	4089	1019	448	0.9	656	89
Crop									
Soybean	20 c	20 b	250	4839	2182 a	880 a	1.5 b	1350 a	161 a
Cowpea	37 a	30 a	193	3902	19 b	10 b	4.0 a	21 b	65 b
Lablab	26 b	32 a	268	4623	0	0	0	0	0
Significance					<u>$P > F$</u>				
Leafing (L)	ns¶	ns	**	ns	ns	ns	ns	ns	ns
Crop (C)	***	**	ns	ns	***	***	*	***	***
L × C	ns	ns	ns	**	ns	ns	ns	ns	ns

* Significant at P = 0.05.

** Significant at P = 0.01.

*** Significant at P = 0.001.

† Vegetative Stage 4.

‡ Reproductive Stage 8.

§ Values followed by different letters within a column are significantly different at P = 0.05 by the least square means test.

¶ Not significant.

Table 4. Leaf concentration of crude protein (CP), P, K, Mg, Ca, Mn, Fe, Cu, Zn, S, and nitrate for P92Y82 soybean, Rongai lablab, and Iron and Clay cowpea at three leaf removal rates, Ames, IA, 2013.

Treatment	Leaf CP	P	K	Mg	Ca	Mn	Fe	Cu	Zn	NO_3–N	S
	g kg^{-1}	mg kg^{-1}									
Leaf removal											
33	256	4,116	24,130	5,264	22,520 b†	94	155	18	128	694	3,550
66	251	4,057	22,760	5,861	25,330 ab	106	293	19	136	515	3,659
99	242	4,198	24,690	5,706	26,870 a	128	210	19	151	553	3,702
Crop											
Soybean	267 a	3,752 c	24,430	6,226 a	16,670 c	74 b	271	17	153	690 a	3,231 c
Cowpea	247 b	4,094 b	23,850	6,144 a	31,410 a	176 a	142	16	142	762 a	3,523 b
Lablab	236 b	4,524 a	23,300	4,461 b	26,640 b	78 b	246	22	121	311 b	4,158 a
Significance						$P > F$					
Leafing (L)	ns‡	ns	ns	ns	*	ns	ns	ns	ns	ns	ns
Crop (C)	**	***	ns	***	***	***	ns	ns	ns	***	***
L × C	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns

* Significant at $P = 0.05$.

** Significant at $P = 0.01$.

*** Significant at $P = 0.001$.

† Values followed by different letters within a column are significantly different at $P = 0.05$ by the least square means test.

‡ Not significant.

concentration differed among crops; cowpea leaves had the greatest concentration, whereas soybean leaves had the least. Cowpea leaves had 47 and 15% greater Ca than soybean and lablab leaves, respectively. Plants at the 99% leaf removal rate had the greatest concentration of Ca in leaves, whereas plots at 33% leaf removal had the lowest. Leaves from plots at 99% leaf removal had 6 and 16% greater Ca than leaves from plots at 66 and 33% leaf removal (Table 4). The concentration of Mn in soybean and lablab leaves did not statistically differ, but both differed from cowpea. Cowpea leaves had 58 and 56% greater Mn than soybean and lablab leaves, respectively. The concentration of S differed by crop (Table 4). Lablab leaves had the greatest concentration of S. This was 15 and 22% greater than that in cowpea and soybean leaves, respectively. The concentrations of K, Fe, Cu, and Zn did not differ by crop or leaf removal rate. Nitrate concentration was greatest in soybean and cowpea leaves and lowest in lablab (Table 4).

Fiber concentrations of harvested leaves differed by crop (Table 6). However, neither leaf removal rate nor the leaf removal rate × crop interaction influenced NDF, ADF, acid detergent lignin (ADL), hemicellulose, cellulose, or ash concentration (Table 6). The concentrations of NDF, ADF, and hemicellulose were greater in soybean and lablab leaves and lowest in cowpea. The concentration of cellulose was greatest in soybean, followed by lablab leaves and lowest in cowpea leaves (Table 6). Soybean leaves had 4 and 8% greater cellulose than lablab and cowpea leaves, respectively. The concentration of ash was not influenced by leaf removal rate or the leaf removal rate × crop interaction (Table 6).

Grain Yield, Aboveground Biomass, and Stand Density, 2014

The interaction of leaf removal rate × crop was not significant for plant stand, aboveground biomass, seed yield, pods m^{-2}, seeds pod^{-1}, and seed weight (Table 7). Stand density at R8 did not differ by rate of leaf harvesting. However, stand density earlier at V4 (before leaf harvesting was done at V6) differed among the crops. The stand densities of CA46 and Top Crop cowpea were similar, and both were greater than that of lablab and soybean (Table 7).

Aboveground biomass varied by entry but not for leaf harvest rate or the leaf harvest rate × crop interaction (Table 7). Soybean had 27, 45, and 46% greater biomass than CA46 cowpea, Top Crop cowpea, and lablab. Aboveground biomass was lowest with Top Crop cowpea and lablab (Table 7).

Plants that did not have leaves removed had the greatest grain yield, whereas plants at 66 and 99% leaf removal yielded the least (Table 7). Plants at 0% leaf removal had 20, 32, and 35% more grain yield than those subjected to 33, 66, and 99% leaf removal, respectively. Grain yield also differed significantly by entry. Soybean had 64 and 72% greater yield than CA46 and Top Crop cowpea, respectively. Leaf removal did not influence pod density (Table 7). Regression analysis showed that grain yield in 2014 decreased as leaf removal rate increased ($r^2 = 0.952$) (Table 5). However, the number of pods m^{-2} differed by crop. Soybean had 80 and 79% more pods than CA46 and Top Crop cowpea, respectively. Pod density was similar among the two cowpea cultivars and averaged 158 m^{-2} (Table 7).

The number of seeds pod^{-1} did not differ significantly for leaf removal rate; however, seed per pod differed among crops; the leaf removal rate × crop interaction was not significant (Table 7). The number of seeds pod^{-1} for CA46 and Top Crop cowpea were similar,

Table 5. Linear functions for leaf removal rate predicting mean pulse leaf Ca, Fe, and Mn concentration, mean yield, mean seed number per square meter and mean seed weight for P92Y82 soybean, Rongai lablab, Iron and Clay cowpea, Top Crop cowpea, and CA46 cowpea in 2013 and 2014.

Parameter	Function	r^2	Year
Ca concentration, mg kg^{-1}	$6895.5x + 20301$	0.963	2013†
Fe concentration, mg kg^{-1}	$2.86x + 55.9$	0.927	2014‡
Mn concentration, mg kg^{-1}	$0.953x + 57.4$	0.952	2014
Mean yield, kg ha^{-1}	$-732.69x + 2116.3$	0.952	2014
Mean seed no., no. m^{-2}	$-369.77x + 1296.2$	0.919	2014
Mean seed wt., mg seed^{-1}	$-16.779x + 154.91$	0.991	2014

† P92Y82 soybean, Rongai lablab, and Iron and Clay cowpea were used for the experiment in 2013.

‡ P92Y82 soybean, Rongai lablab, Top Crop cowpea, and CA46 cowpea were used for the experiment in 2014.

whereas soybean had the lowest number of seeds pod^{-1}. CA46 cowpea had 53% more seeds pod^{-1} than soybean, whereas Top Crop cowpea had 43% more seeds pod^{-1} than soybean. The number of seeds m^{-2} was influenced by leaf removal rate and crop, but not by their interaction (Table 7). Regression analysis showed that the average number of seeds m^{-2} decreased with increase in leaf removal rate (r^2 = 0.919) (Table 5). Soybean yielded the most seeds m^{-2}. The number of seeds m^{-2} for the two cowpea cultivars was similar. The no-leaf removal control yielded more seeds than plants with leaves removed (Table 7). Plants at 0% leaf removal had 17% more seeds m^{-2} than those with 33% leaf removal, and 28% more seeds m^{-2} than those at 66 and 99% leaf removal. Seed weight also differed by crop but not by leaf removal rate or the leaf removal rate × crop interaction (Table 7). Regression analysis showed that average seed weight decreased as leaf removal rate increased (r^2 = 0.991) (Table 5). Soybean seeds weighed 21 and 29% more than CA46 and Top Crop

Table 6. Neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL), hemicellulose, cellulose, and ash concentrations of leaves for P92Y82 soybean, Iron and Clay cowpea, and Rongai lablab, Ames, IA, 2013.

Treatment	NDF	ADF	ADL	Hemicellulose	Cellulose	Ash
	g kg^{-1}					
Leafing						
33	413	316	64.8	97.6	251	5.2
66	406	308	64.9	96.8	243	4.9
99	404	308	64.5	97.9	244	5.3
Crop						
Soybean	425 a†	327 a	71 a	98 a	256 a	5.7
Cowpea	383 b	292 b	56 b	91 b	235 b	4.9
Lablab	417 a	313 a	67 a	103 a	246 ab	4.9
Significance			$P > F$			
Leafing (L)	ns‡	ns	ns	ns	ns	ns
Crop (C)	***	**	***	***	*	ns
L × C	ns	ns	ns	ns	ns	ns

* Significant at P = 0.05.

** Significant at P = 0.01.

*** Significant at P = 0.001.

† Values followed by different letters within a column are significantly different at P = 0.05 by the least square means test.

‡ Not significant.

Table 7. Crop stand density, aboveground biomass and yield components for P92Y82 soybean, Rongai lablab, CA46 cowpea, and Top Crop cowpea, Ames, IA, 2014.

Treatment	Stand density V4	Stand density R8	Dry leaves	Biomass	Yield	Pods	Seeds		Seed wt.	Seed CP
	no. m^{-2}		kg ha^{-1}			no. m^{-2}	no. pod^{-1}	no. m^{-2}	mg seed^{-1}	g kg^{-1}
Leaf removal										
0	37	38	0	5007	2375 a†	332	4.8	1439 a	160 a	292
33	36	35	122 b	4111	1909 ab	410	3.8	1190 ab	151 ab	286
66	36	37	435 a	4896	1616 b	344	4.2	1042 b	143 b	284
99	36	35	451 a	4737	1548 b	317	4.6	1036 b	141 b	296
Crop										
Soybean	32 b	42	325 ab	6673 a	3411 a	743 a	2.7 b	1943 a	178 a	353 a
CA46	38 a	34	422 a	4847 ab	1220 b	151 b	5.8 a	857 b	141 b	266 b
Top Crop	41 a	33	359 a	3646 b	955 b	158 b	4.7 a	731 b	127 c	248 c
Lablab	33 b	–	239 b	3586 b	0	–	–	–	–	–
Significance					$P > F$					
Leafing (L)	ns‡	ns	***	ns	**	ns	ns	**	**	ns
Crop (C)	***	ns	*	**	***	***	***	***	***	***
L × C	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns

* Significant at P = 0.05.

** Significant at P = 0.01.

*** Significant at P = 0.001.

† Values followed by different letters within a column are significantly different at P = 0.05 by the least square means test.

‡ Not significant.

cowpea seeds, respectively (Table 7). Soybean had the greatest CP concentration, followed by CA46 and Top Crop cowpea with the lowest concentrations (Table 7). Soybean seeds had 25 and 30% more CP than seed of CA46 and Top Crop cowpea. Seed weight differed by leaf removal rate and crop, but not for their interaction (Table 7). Seeds from plants at 0% leaf removal weighed more than seeds from plots at 33, 66, and 99% leaf removal. Seeds from plots that were not leafed weighed 7, 11, and 12% more than seeds from plots at 33, 66, and 99% leaf removal, respectively.

Nutrient and Fiber Concentrations in Harvested Leaves, 2014

Leaf removal rate and crop influenced Mn concentration of harvested leaves but the interaction of leaf removal rate × crop did not (Table 8). The greatest concentration of Mn in harvested leaves occurred at the 99% removal rate (Table 8). The concentration of Mn at 99% leaf removal was 31 and 47% more than that 66 and 33% leaf removal, respectively. Top Crop and CA46 cowpeas had the greatest Mn concentration and did not differ from each other; soybean and lablab had the lowest Mn concentration in removed leaves. The leaves from CA46 cowpea had about 50% more Mn than soybean and lablab, respectively, whereas Top Crop cowpea had about 41% more Mn than soybean and lablab leaves, respectively (Table 8). Overall, regression analysis showed that leaf Mn concentration increased with increase in leaf removal rate ($r^2 = 0.952$) (Table 5). The concentration of Fe in harvested leaves was influenced by crop (Table 8) and regression analysis showed that Fe concentration increased with increase in leaf removal rate ($r^2 = 0.927$) (Table 5). Leaf removal at 99% resulted in 38 and 65% greater Fe concentration than 66 and 33% leaf removal. Lablab leaves had the greatest Fe concentration, whereas soybean and Top Crop cowpea had the lowest. Lablab leaves had 33, 42, and 50% greater Fe than CA46 cowpea, soybean, and Top Crop cowpea leaves, respectively (Table 8). The concentrations of Mg and Ca were not influenced by leaf removal rate; however, these nutrients differed significantly among crops (Table 8). The concentration of Mg was greatest in soybean and CA46 cowpea leaves and lowest in lablab. Soybean leaves had 19 and 51% greater Mg than Top Crop cowpea and lablab leaves, whereas CA46 cowpea leaves had 8 and 44% greater Mg than Top Crop cowpea and lablab leaves, respectively (Table 8). Calcium concentration was greatest in Top Crop cowpea leaves and lowest in soybean leaves. Top Crop cowpea leaves had 23, 24, and 32% greater Ca than CA46 cowpea, lablab, and soybean, respectively (Table 8). Leaf removal rate, crop, and the leaf removal rate × crop interaction did not impact concentrations of P, K, Cu, Zn, S, or CP (Table 8). Nitrate concentration in harvested leaves was influenced by crop but the leaf removal rate and leaf removal rate × crop interaction did not (Table 8). Nitrate concentration was greatest in CA46 cowpea leaves and lowest in lablab, whereas soybean and Top Crop cowpea were intermediate (Table 8).

The concentration of ADF in harvested leaves was influenced by crop, but the effect of leaf harvest percentage × crop interaction was not significant (Table 9). Soybean had the greatest ADF concentration in harvested leaves whereas lablab had the lowest; the ADF concentration in leaves was similar for the two cowpea entries. Soybean leaves had 25 and 27% more ADF than the two cowpea cultuvars and lablab, respectively (Table 9). The concentration of ADL was influenced by leaf harvest rate and crop, but the interaction of leaf harvesting rate × crop was not significant (Table 9). Leaves from the 66% removal rate had 3 and 26% more ADL than leaf removal at 99 and 33%. Soybean leaves also had the greatest amount of ADL, whereas CA46 cowpea had the lowest. The ADL concentration in Top Crop cowpea and lablab leaves were not different. Soybean leaves had 24 and 11% more ADL than CA46 cowpea, Top Crop cowpea, and lablab, respectively. Neither leaf removal rate nor grain legume entry influenced concentrations of NDF, hemicellulose or dry matter concentration of harvested leaves in 2014 (Table 9).

Cellulose concentration differed by crop but the leaf removal rate × crop interaction was not significant (Table 9). Mean cellulose

Table 8. Leaf concentration of crude protein (CP), nitrate, P, K, Mg, Ca, Mn, Fe, Cu, Zn, and S for P92Y82 soybean, Rongai lablab, CA46 cowpea, and Top Crop cowpea at three leaf removal rates, Ames, IA, 2014.

Treatment	Leaf CP	P	K	Mg	Ca	Mn	Fe	Cu	Zn	NO_3–N	S
	g kg^{-1}	mg kg^{-1}									
Leaf removal											
33	223	3,889	21,790	3,705	10,750	83 b†	104 b	10	56	291	2,380
66	213	3,332	21,290	3,501	12,180	107 b	184 b	9	53	282	2,233
99	203	2,936	18,690	3,591	14,650	156 a	297 a	9	29	206	2,083
Crop											
P92Y82	234	2,781	22,470	4,505 a	10,640 b	81 b	165 b	8	32	288 ab	2,159
CA46	243	3,432	22,030	3,999 a	12,070 ab	163 a	189 ab	10	44	434 a	2,211
Top Crop	207	3,411	20,040	3,667 ab	15,600 a	138 a	141 b	10	59	185 ab	2,203
Rongai	169	3,919	17,820	2,225 b	11,790 ab	80 b	284 a	9	48	131 b	2,159
Significance						$P > F$					
Leafing (L)	ns‡	ns	ns	ns	ns	**	***	ns	ns	ns	ns
Crop (C)	ns	ns	ns	**	*	***	**	ns	ns	*	ns
L × C	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns	ns

* Significant at $P = 0.05$.

** Significant at $P = 0.01$.

*** Significant at $P = 0.001$.

† Values followed by different letters within a column are significantly different at $P = 0.05$ by the least square means test.

‡ Not significant.

Table 9. Neutral detergent fiber (NDF), acid detergent fiber (ADF), acid detergent lignin (ADL), hemicellulose, cellulose, and ash concentrations from leaves for P92Y82 soybean, CA46 cowpea, Top Crop cowpea, and Rongai lablab, Ames, IA, 2014.

Treatment	NDF	ADF	ADL	Hemicellulose	Cellulose	Ash
	g kg^{-1}					
Leaf removal						
33	201	153	26 b†	48	127	0.3 b
66	237	177	35 a	60	143	3.0 a
99	217	159	34 ab	58	125	4.8 a
Crop						
Soybean	264	202 a	37 a	61	165 a	2.2 b
CA46	208	151 b	28 b	57	124 b	1.6 b
Top Crop	207	151 b	28 ab	56	123 b	1.1 b
Lablab	195	148 c	33 ab	47	115 c	6.0 a
Significance			$P > F$			
Leafing (L)	ns‡	ns	**	ns	ns	**
Crop (C)	ns	*	***	ns	*	***
L × C	ns	ns	ns	ns	ns	ns

* Significant at $P = 0.05$.

** Significant at $P = 0.01$.

*** Significant at $P = 0.001$.

† Values followed by different letters within a column are significantly different at $P = 0.05$ by the least square means test.

‡ Not significant.

concentration across leaf removal rates was 132 g kg^{-1}. Leaves from soybean had the greatest concentration of cellulose and lablab had the lowest. Soybean leaves had 25% more cellulose than CA46 and Top Crop cowpea and 30% more than lablab leaves. Harvested leaf ADL concentration differed by leaf removal rate and by crop. However, the influence of leaf removal rate × crop interaction was not significant for ADL concentration (Table 9). Soybean had the greatest concentration of ADL with 24% more ADL than CA46 and Top Crop cowpea, and 8% more than lablab. A leaf harvesting percentage of 66% resulted in the greatest leaf ADL concentration, whereas 33% resulted in the lowest. ADL leaf concentration at 66% leaf harvest was 3 and 26% more than that at 99 and 33%.

Harvested leaf ash concentration varied by leaf removal rate and crop, but not their interaction (Table 9). Leaves from plants with 66 and 99% leaf removal rates had the greatest concentration of ash, whereas those at 33% leaf removal had the lowest. Lablab leaves had 82, 73, and 63% more ash than Top Crop cowpea, CA46 cowpea, and soybean leaves, respectively (Table 9).

DISCUSSION

Impact of Leaf Removal on Aboveground and Yield Components

While assessing the impact of leaf removal on aboveground biomass and other yield components, the greater soybean aboveground biomass that was found compared to cowpea and lablab corresponds with Rao and Northup (2009), who reported that soybean produced more biomass than cowpea for 3 yr in a 4-yr experimental study in Oklahoma, USA. The amount of cowpea biomass in our study also corresponds with previous studies by Agza et al. (2012) who reported that aboveground biomass accumulation of different cowpea genotypes ranged from 2330 to 7670 kg ha^{-1}.

Rongai lablab is a short-day plant that is quite sensitive to day length and flowers best with less than 11 h of daylight, although it is reported to require ample sunlight. Day length in Ames, IA, was between 16 to 12 h during the 2013 and 2014 growing seasons. Rongai lablab usually seeds late and has low frost tolerance (FAO, 2015). Greater soybean yield compared to the three cowpea cultivars in both 2013 and 2014 and may be attributable to soybean breeding efforts done over many decades in the midwestern United States, whereas no breeding efforts have been made to improve cowpea or lablab in this region. Some of those soybean breeding efforts include identification and characterization of major genes and quantitative trait loci (QTL). These, for instance, influence soybean maturity group and its (maturity group) impact on leaves, signals to start or delay flowering among others, which in turn influence soybean yield compared with other legumes in the study (Mourtzinis and Conley, 2017). Decreased seed yields following leaf removal from cowpea and soybean may have resulted from a reduction in source leaves that limited the reproductive sink size (Bubenheim et al., 1990). Hoogesteger and Karlsson (1992) reported that leaf removal altered photosynthesis directly through changing source–sink relations. They explained that when leaves were harvested, photosynthates were directed toward the development of new leaves at the expense of being used for grain production. Greater soybean grain yield compared with cowpeas grain yield may be attributed to differences in variety and genetic makeup of the crops. In 2013, Iron and Clay cowpea produced very few seeds of very small size; therefore, analysis for CP was not done that year to determine the effects of leaf removal among soybean and Iron and Clay cowpea. In 2014, greater soybean seed CP concentration than CA46 and Top Crop cowpea seeds may be explained by a decline in cowpea CP as the plants mature. Awolumate (1983) explained that, although cowpea accumulated N at a rate much faster than soybean during seed development, 994 µg d^{-1} compared with 473 µg d^{-1}, respectively, cowpea biomass CP decreased with development time. He reported that cowpea biomass CP decreased from 40% in early seed development stages to 26% in mature cowpea compared with decrease from 35 to 33% in soybean. This decrease in cowpea biomass CP toward maturity could explain why soybean seeds had more CP than cowpea seeds. The nonsignificant effects of leaf removal rate on seed CP in 2014 are in line with findings by Burton et al. (1995) and Lawn and Brun (1974), who reported seed protein concentration was not influenced by leaf removal. Decreased seed CP with leaf removal is attributed to the reduction in vegetative N remobilization, which causes lower seed protein concentrations (Burton et al., 1995). Nonsignificant effects of leaf removal on seed protein may be explained by compensatory regrowth, which then facilitates vegetative N remobilization for seed protein.

Impact of Leaf Removal on Legume Leaf Nutrients

Grain legume leaves contained considerable amounts of nutrients, including several vital micronutrients such as Fe and Zn, which are commonly deficient in human diets in developing countries. Although leaf removal rate did not significantly affect several of the nutrient concentrations in 2013 or 2014, the lower leaf concentration of Zn observed in 2014 compared with 2013 may be attributable to lower soil Zn levels at the 2014 experimental site (Alloway, 2008). The increase in harvested leaf Ca, Fe, and Mn concentrations with increased leaf removal rate may likely be attributable to nonmobility of these elements. Elemental nonmobility may explain their lower concentrations in newer leaves/tissues and greater concentration in older leaves (Owen and Kissel, 2015). This may explain why the higher percentage of leaf removal from top (younger leaves) moving down to older leaves resulted in higher concentration of these nutrients at 99 and 66% leaf removal compared with 33% leaf removal.

The influence of crop species and growing environment on most of the leaf nutrient concentrations is evident in 2013 and 2014 results despite the use of different cowpea cultivars each year and separate data analysis for both years. Similarly, Oelberg (1956) elaborated that differences in leaf nutritive value were attributable to plant species. For instance, legume leaves had more Ca than grass leaves (Oelberg, 1956). Although other factors such as climate and soil factors may have influenced leaf nutrient composition, plant species had the greatest influence on leaf nutritional composition in our study. Fiber differences among crops may be attributable to differences in cell and wall development of these legumes including small differences in amounts of phenolic acids (Allen and Jung, 2014). Unlike grasses whose digestibility decreases rapidly, legume leaf digestibility changes slightly over time with age and maturity (Buxton, 1996). This may explain why we did not observe much effect of leaf removal rates on fiber concentrations in 2013 and only for ADL and ash in 2014. A decrease in leaf/stem ratio as plants advance in maturity, along with increase in leaf cell-wall concentration and decreased cell solubles may explain why we observed greater concentrations of ADL and ash in 2014 with greater leaf removal rates (older leaves at the bottom of the plant closer to the ground) (Buxton, 1996). Although comparative differences for leaf nutritive value existed among entries, all the grain legume leaves would be excellent supplements, especially during dry seasons when forage availability is low, and livestock is surviving on low quality feeds.

A slightly greater soybean leaf CP concentration (264 g kg^{-1}) in our study compared with some previous studies may be attributable to differences in the stage at which the leaves were harvested. For instance, Blount et al. (2009) reported soybean leaf CP of 178 g kg^{-1} when leaves were picked at 50% bloom. Lower CP in lablab leaves compared with soybean could be attributable to poor rhizobia infection and subsequent low or nonexistent N fixation (Bulyaba and Lenssen, 2017). The authors reported no nodulation of lablab without rhizobium inoculation.

CONCLUSION

The utilization of soybean, cowpea, and lablab leaves as leafy vegetables or forage may improve human and ruminant nutrition by putting to use leaves that would otherwise be left in the field. For humans, these grain legume leaves can be an important source of Fe, Zn, and dietary fiber. Additionally, the leaves are an excellent supplement for protein and other nutrients for livestock during the dry season. It is also important to note that the amount of micronutrients available in harvested grain legume leaves may differ relative to available soil concentrations and our results may not be indicative of concentrations from plants grown on degraded Ferralsols or Oxisols. Our experiment, therefore, provides a baseline for nutrient concentration of grain legume leaves for soybean, cowpea, and lablab, which could be compared with subsequent studies conducted on lower fertility soils. With our research findings, we would recommend leaf removal/harvest of at least two leaves from the grain legume crops at V6 to utilize as leafy vegetables or forage without negatively affecting grain yield. How the crops recover from leaf removal may be influenced by the presence or absence of environmental stresses such as drought or soil infertility. In effort to overcome micronutrient deficiency especially Fe and Zn, lablab leaves would be most recommended. Given a 2-yr average, lablab leaves numerically appeared to have the greatest concentration of those two nutrients. Supplementary nutritional analysis of the leaves is necessary to determine the amount and availability of other vital nutrients such as folate. There is also need to study how different soils influence leaf nutritive value of these grain legumes. Additionally, efficient leaf harvesting methodology is necessary and would be valuable to aid the leaf removal process while minimizing crop injury.

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REFERENCES

Agza, B., K. Binyam, Z. Solomon, A. Eskinde, and A. Ferede. 2012. Animal feed potential and adaptability of some cowpea (*Vigna unguiculata*) varieties in northwest lowlands of Ethiopia. J. Agric. Res. 11:478–483.

Allen, M.S., and H.G. Jung. 2014. Characteristics of plant cell walls affecting intake and digestibility of forages by ruminants. J. Anim. Sci. 73:2774–2790.

Alloway, B.J. 2008. Zinc in soil and crop nutrition. 2nd edition. International Zinc Association (IZA) and International Fertilizer Association, Brussels, Belgium and Paris, France.

Awolumate, E. 1983. Accumulation and quality of storage protein in developing cowpea, mung bean and soya bean seeds. J. Sci. Food Agric. 34:1351–1357. doi:10.1002/jsfa.2740341206

Badi, S.H., H.D. Dikwahal, and G.G. Jibung. 2012. Response of vegetable cowpea (*Vigna unguiculata* (L.) Walp.) to intra-row spacing and defoliation at Garkawa. Asian J. Agric. Sci. 4:210–212.

Baloyi, B.M., and V.I. Ayodele. 2013. Effects of leaf harvest on crude protein and mineral contents of selected early maturing lines of lablab (*Lablab purpureus*). Afr. J. Agric. Res. 8:449–453.

Barrett, R.P. 1987. Integrating leaf and seed production strategies for cowpea (*Vigna unguiculata* (L.) Walp.). M.S. thesis. Michigan State Univ., East Lansing.

Barrett, R.P. 1990. Legume species as leaf vegetables. In: J. Janick and J.E. Simon, editors, Advances in new crops. Timber Press, Portland, OR. p. 391–396.

Blount, A.R.S., D.L. Wright, R.K. Sprenkel, T.D. Hewitt, and R.O. Myer. 2009. Forage soybeans for grazing, hay and silage. Publ. SS-AGR-180. University of Florida, IFAS Extension, Gainesville, FL. https://edis.ifas.ufl.edu/ag184 (accessed 27 Nov. 2017).

Board, J.E., and B.G. Harville. 1993. Soybean yield component responses to a light interception gradient during reproductive period. Crop Sci. 33:772–777. doi:10.2135/cropsci1993.0011183X003300040028x

Board, J.E., A.T. Wier, and D.J. Boethel. 1994. Soybean yield reductions caused by defoliation during mid to late seed filling. Agron. J. 86:1074–1079. doi:10.2134/agronj1994.00021962008600060027x

Bremmer, J.M. 1996. Nitrogen—total. In: D.L. Sparks, A.L. Page, P.A. Helmke, and R.H. Loeppert, editors, Methods of soil analysis, Part 3. SSSA Book Ser. 5.3. SSSA and ASA, Madison, WI. p. 1085–1121. doi:10.2136/sssabookser5.3.c37

Bubenheim, D.L., C.A. Mitchell, and S.S. Nielsen. 1990. Utility of cowpea foliage in crop production for space. In: J. Janick and J.E. Simon, editors, Advances in new crops. Timber Press, Portland, OR. p. 535–538.

Bulyaba, R., and A.W. Lenssen. 2017. Influence of *Bradyrhizobium* inoculation and fungicide seed treatment on development and yield of cowpea, lablab and soybean. Crop Forage Turfgrass Manage. 3. doi:10.2134/cftm2017.01.0007

Burton, J.W., D.W. Israel, R.F. Wilson, and T.E. Carter. 1995. Effects of defoliation on seed protein concentration in normal and high protein lines of soybean. Plant Soil 172:131–139. doi:10.1007/BF00020867

Buxton, D.R. 1996. Quality-related characteristics of forages as influence by plant environment and agronomic factors. Field Crops Res. 59:37–49.

Combs, S.M., and M.V. Nathan. 1998. Soil organic matter. p. 53–58. In: J.R. Brown, editor, Recommended chemical soil test procedures for the North Central region. North Central Regional Res. Publ. 221. Missouri Agric. Exp. Stn., Univ. of Missouri, Columbia, MO.

Demooy, B.E., and C.J. Demooy. 1989. Effects of leaf-harvesting practices on yield and yield components of ER-7 cowpea (*Vigna unguiculata* (L.) Walp.) in semi-arid Botswana. Field Crops Res. 22:27–31. doi:10.1016/0378-4290(89)90086-5

Egli, D.B., and J.E. Leggett. 1976. Rate of dry matter accumulation in soybean seeds with varying source-sink ratios. Agron. J. 68:371–374. doi:10.2134/agronj1976.00021962006800020042x

Enyi, B.A.C. 1975. Effects of defoliation on growth and yield in groundnut (*Arachis hypogea*), cowpeas (*Vigna unguiculata*), soybean (*Glycine max*) and green gram (*Vigna aurens*). Ann. Appl. Biol. 79:55–66. doi:10.1111/j.1744-7348.1975.tb01522.x

Fehr, W.R., B.K. Lawrence, and T.A. Thompson. 1981. Critical stages of development for defoliation of soybean. Crop Sci. 21:259–262. doi:10.2135/cropsci1981.0011183X002100020014x

Food and Agriculture Organization of the United Nations (FAO). 2004. Monitoring progress towards the World Food Summit and Millinnium Development Goals. FAO, Rome. p 6–17.

Food and Agriculture Organization of the United Nations (FAO). 2015. *Lablab purpureus* (L.) Sweet. FAO, Rome. http://www.fao.org/ag/agp/agpc/doc/gbase/data/pf000047.html (accessed 27 Nov. 2017).

Gelderman, R.H., and D. Beegle. 1998. Nitrate-nitrogen. p. 17–20. *In* Recommended chemical soil test procedures for the North Central Region. North Central Regional Res. Publ. 221. Missouri Agric. Exp. Stn., Univ. of Missouri, Columbia, MO.

Gibson, R.M., R.L. Lovvorn,, and B.E.W. Smith. 1943. Response of soybeans to experimental defoliation. J. Am. Soc. Agron. 35:768–778. doi:10.2134/agronj1943.00021962003500090003x

Goli, A., and D.B. Weaver. 1986. Defoliation responses of determinate and indeterminate late planted soybeans. Crop Sci. 26:156–166. doi:10.2135/cropsci1986.0011183X002600010036x

Hofstrand, J. 2010. Economics of tile drainage. Iowa State Univ., Ames. https://www.extension.iastate.edu/agdm/articles/hof/HofJuly10.html (accessed 24 Dec. 2018).

Hoogesteger, J., and P.S. Karlsson. 1992. Effects of defoliation on radial stem growth and photosynthesis in the mountain birch (*Betula pubescens* spp. tortuosa). Funct. Ecol. 6:317–323.

Horneck, D.A., and R.O. Miller. 1998. Determination of total nitrogen in plant tissue. In: Y.P. Karla, editor, Handbook of reference methods for plants analysis. CRC Press, Boca Raton, FL.

Johnston, T.J., and J.W. Pendleton. 1968. Contribution of leaves at different canopy levels to seed production of upright and lodged soybeans (*Glycine max* (L.) Merrill). Crop Sci. 8:291–292. doi:10.2135/cropsci1968.0011183X00080003009x

Karikari, S.K., and G. Molatakgosi. 1999. Response of cowpea [*Vigna unguiculata* (L.) Walp.] varieties to leaf harvesting in Botswana. UNISWA J. Agric. 8:5–11.

Lawn, R.J., and W.A. Brun. 1974. Symbiotic nitrogen fixation in soybeans: I. Effect of photosynthetic source sink manipulations. Crop Sci. 14:11–16. doi:10.2135/cropsci1974.0011183X001400010004x

Licht, M. 2014. Soybean growth and development. Publ. PM 1945. Iowa State University Coop. Ext. Service, Ames, IA.

Mallarino, A., J.E. Sawyer, and S.K. Barnhart. 2013. A general guide for crop nutrient and limestone recommendations in Iowa. Publ. PM 1688. Iowa State University Coop. Ext. Service, Ames, IA.

Mourtzinis, S., and S.P. Conley. 2017. Delineating soybean maturity groups across the United States. Agron. J. 109:1397–1403. doi:10.2134/agronj2016.10.0581

Muller, O., and M. Krawinkel. 2005. Malnutrition and health in developing countries. Can. Med. Assoc. J. 173:279–286. doi:10.1503/cmaj.050342

Mulvaney, R.L. 1996. Nitrogen—inorganic forms. In: D.L. Sparks, A.L. Page, P.A. Helmke, and R.H. Loeppert, editors, Methods of soil analysis, Part 3. SSSA Book Ser. 5.3. SSSA and ASA, Madison, WI. p. 1123–1184. doi:10.2136/sssabookser5.3.c38

National Cooperative Soil Survey (NCSS). 2015a. Canisteo series. USDA, Washington, DC. https://soilseries.sc.egov.usda.gov/OSD_Docs/C/CANISTEO.html (accessed 27 Nov. 2017).

National Cooperative Soil Survey (NCSS). 2015b. Clarion series. USDA, Washington, DC. https://soilseries.sc.egov.usda.gov/OSD_Docs/C/CLARION.html (accessed 27 Nov. 2017).

National Cooperative Soil Survey (NCSS). 2015c. Nicollet series. USDA, Washington, DC. https://soilseries.sc.egov.usda.gov/OSD_Docs/N/NICOLLET.html (accessed 27 Nov. 2017).

Natural Resources Conservation Service (NRCS). 2014. NRCS-MLRA 103 Soil survey project. USDA, Washington, DC. https://www.nrcs.usda.gov/wps/portal/nrcs/detail/mn/newsroom/?cid=stelprdb1242954 (accessed 26 Oct. 2017).

Oelberg, K. 1956. Factors affecting the nutritive value of range forage. J. Range Manage. 9:220–225. doi:10.2307/3894056

Ogedegbe, S.A., V.B. Ongunlela, O.O. Olufajo, and E.C. Odion. 2012. Seed yield and yield attributes of lablab as influenced by phosphorous application, cutting height. Asian. J. Crop. Sci. 4:12–22.

Owen, C.P., and D.E. Kissel. 2015. Plant analysis handbook for Georgia. University of Georgia, Athens. http://aesl.ces.uga.edu/publications/plant/Nutrient.asp (accessed 18 Sept. 2017).

Pella, E. 1990. Elemental organic analysis. Part 1. Am. Lab. 22:116–125.

Rao, S.C., and B.K. Northup. 2009. Capabilities of four novel warm-season legumes in the southern great plains: Biomass and forage quality. Crop Sci. 49:1096–1102. doi:10.2135/cropsci2008.08.0499

Saidi, M., M. Ngouajio, F.M. Itulya, and J. Ehlers. 2007. Leaf harvesting initiation time and frequency affect biomass partitioning and yield of cowpea. Crop Sci. 47:1159–1166. doi:10.2135/cropsci2006.06.0420

Schwartz, H.F., and M.A.C. Langham. 2010. Cowpea growth stages. Colorado State Univ., Fort Collins, and South Dakota State Univ., Brookings. https://beanipm.pbgworks.org/cowpea (accessed 8 Nov. 2017).

Teigen, J.B., and J.J. Vorst. 1975. Soybean response to stand reduction and defoliation. Agron. J. 67:813–816. doi:10.2134/agronj1975.00021962006700060022x

USEPA. 1986. Test methods for evaluating solid waste. Vol. IA. 3rd ed. EPA/SW-846. National Technical Information Service, Springfield, VA.

Warncke, D., and J.R. Brown. 1998. Potassium and other cations. p. 31–33. In: J.R. Brown et al., editors, Recommeded chemical soil test procedures for the North Central Region. North Central Regional Res. Publ. 221. SB 1001. Missouri Agric. Exp. Stn., Columbia, MO.

Watson, M.E., and J.R. Brown. 1998. pH and lime requirement. p. 13–16. In: J.R. Brown et al., editors. Recommended chemical soil test procedures for the North Central Region. North Central Regional Res. Publ. 221. SB 1001. Missouri Agric. Exp. Stn., Columbia, MO.

Weber, C.R. 1955. Effects of defoliation and topping simulation hail injury to soybeans. Agron. J. 47:262–266. doi:10.2134/agronj1955.00021962004700060007x

Wood, I. 1983. Lablab bean (*Lablab purpureus*) for grain and forage production in the Ord irrigation area. Aust. J. Exp. Agric. 23:162–171. doi:10.1071/EA9830162

World Health Organization. 2017. Nutritional anaemias. Tools for effective prevention and control. World Health Organization, Geneva, Switzerland. http://apps.who.int/iris/bitstream/handle/10665/259425/9789241513067-eng.pdf?sequence=1&ua=1 (accessed 15 Dec. 2018).